NEW DIETARY INGREDIENT NOTIFICATION

Submitted by Greenberg Traurig, LLP, on Behalf of:
Stage II Enterprises, LLC

Dated: February 3, 2004

Contact Person: Jim Prochnow, Attorney at Law
Greenberg Traurig, LLP
The Tabor Center
1200 17th Street, Suite 2400
Denver, CO 80202
T: 303-572-6546
F: 720-904-7646
E: prochnowj@gtlaw.com

Or

Robert McCaleb, Natural Product Specialist
Greenberg Traurig, LLP
The Tabor Center
1200 17th Street, Suite 2400
Denver, CO 80202
T: 303-572-6555
F: 720-904-7655
E: mccalebr@gtlaw.com

We consider all of the information provided in this submission to be confidential, and should not be disclosed to anyone other than employees and agents of the FDA because such information is confidential trade secret information exempt, pursuant to the FOIA, from disclosure. We note that Section 8 of DSHEA, codified as Sec. 413(A) of the FFDCA, does not override or preclude the application of the FOIA. If the FDA or HHS intends to make public any of this information, we request that we be notified of that intent at least 20 calendar days before the proposed publication.

If the FDA objects to this broad, all-inclusive claim of exemption from publication, we shall work with the FDA to arrive at a mutually agreeable decision as to the portions of this notification which may be displayed to the public.

NEW DIETARY INGREDIENT NOTIFICATION

This notification is being submitted on behalf of Stage II Enterprises, LLC ("Stage II"). Stage II submits this New Dietary Ingredient ("NDI") notification for Cotinine pursuant to Section 8 of the Dietary Supplement Health and Education Act of 1994 (codified at §21 U.S.C. § 350b) of the Federal Food, Drug and Cosmetic Act and pursuant to 21 C.F.R. § 190.6. This NDI notification presents the scientific information upon which the Company bases its conclusion that Cotinine will reasonably be expected to be safe for human consumption as an ingredient in a dietary supplement at the consumption level specified below (75-150 mg/day).

The format of this notification follows the regulatory numbering scheme of 21 C.F.R. § 190.6(b)(1-5).

(1) Name and Address of the Manufacturer of the Supplement: The names and address information are not being provided for the manufacturer or distributor of any dietary supplement that will contain Cotinine since there has been no decision yet as to whether this ingredient will be sold, in bulk, by the Company, as an ingredient to a manufacturer or distributor of a dietary supplement, or will be included in a supplement that will be manufactured or distributed or both by the Company. The name of the entity which is submitting this notification and which is responsible for the NDI's production, marketing and sales is:

Stage II Enterprises, LLC
2458 E. Russell Road, Ste. B
Las Vegas, NV 89120

(2) Name of the NDI: (-)-Cotinine. This notification is being filed for two forms of (-)-Cotinine:

(i) A synthetic form: (-)-Cotinine is produced in a laboratory from commercially available chemicals.

[ii] A natural form: A purified form of (-)-Cotinine derived from the plant *Duboisia hopwoodii [Solanaceae]*.

(3) Description of the dietary supplement containing the new dietary supplement ingredient. No such information for a dietary supplement is being provided for the reason stated in para. (1) above. The nature of specific dietary supplements containing the new dietary ingredient that is the subject of this notification will be determined by the entity who purchases and markets this ingredient. The following descriptive information is being provided for this ingredient:

(i) The level of the NDI in any dietary supplement is recommended by the Company to be in the range of 75 mg. to 150 mg. per day.

(ii) The Company expects the NDI to be included in some type of standard edible container such as a capsule or a tablet.

(iii) The chemical structure of synthetic and naturally derived (-)-Cotinine is identical and is depicted and explained at Tab 1. There are two isomers of Cotinine: (-)-cotinine and (+)-cotinine. However, only (-)-cotinine is naturally occurring in *Duboisia hopwoodii*.

(iv) The Company intends to sell (-)-Cotinine for use as a dietary ingredient in dietary supplements. The conditions of use to be suggested on the label are "As a dietary supplement take one (1) to four (4) tablets (or capsules) per day or as directed by a health care provider." The amount per tablet or capsule will be adjusted to keep the daily serving within the range of 75 mg to 150 mg per day.

(4) Safety. The following information consists of the evidence of safety required by 21 C.F.R. § 190.6(b)(4):

(A) Current Relevant Regulatory Provisions. Cotinine exists naturally as a substance in the Australian plant, *Duboisia hopwoodii* at levels equivalent to the intended daily consumption level range described in para. (3)(i) above. *Duboisia hopwoodii* is not listed in *Herbs of Commerce*, nor is it a current dietary supplement ingredient, to the best of our knowledge. *Duboisia hopwoodii* is not the subject of this notification, and the Company does not intend to market this plant; only the purified Cotinine from this plant, or its synthetic equivalent.

(B) History of Safe Use. The Company is not aware of any history of human consumption of (-)-Cotinine, as a sole dietary ingredient, whether extracted from plants or made synthetically.

(C) Studies of Cotinine:

Based on the literature attached, and other studies, we believe that Cotinine can be reasonably expected to be safe for use as a dietary supplement ingredient at the level indicated above. Cotinine has been studied extensively. Its safety has been established by conventional toxicological evaluations, and its physiological and behavioral effects are known from both *in vivo* and clinical

assessments. A brief summary of the findings detailed in the attached literature follows, below:

ABSORPTION AND METABOLISM

1) Terminal elimination half- life = 14-20 hrs (Benowitz et al. 1983)
2) Nearly complete oral bioavailability (DeSchepper et al. 1987)

NEUROPHARMACOLOGY OF COTININE

1) Increased 5-HT synaptic activity (Essman 1973; Fuxe et al. 1979)
2) Poor cholinergic nicotinic receptor affinity (Abood et al. 1981)
3) Mecamylamine without effect (Goldberg et al. 1989)
4) Octanol/water ratio = 100 (Lippiello & Caldwell 1992)
5) Increased dopamine release (Lippiello & Caldwell 1992)
6) Inhibits nicotinic receptor binding at higher concentrations (NovaScreen)

CARDIOVASCULAR EFFECTS

1) IV administration decreases BP in dogs (Borzelleca et al. 1963)
2) Decreased BP in cats (Yamamoto & Domino 1965)
3) Induces intestinal smooth muscle relaxation (Kim et al. 1969)
4) Dose-dependent decreases in HR & BP in rats (Dominiak et al. 1985)
5) Minimal effect in humans (Benowitz et al.1983; DeSchepper et al. 1987)
6) No change in BP, HR & ECG (Scherer et al. 1988; Curvall et al. 1990)
7) Decreased MAP & no effect on ECG in humans (Keenan et al. 1994)

TOXICOLOGY

1) Oral LD50 = 1604 + 452 mg/kg (Borzelleca et al. 1962)
2) Intraperitoneal LD50 = 930 + 35 mg/kg (Borzelleca et al. 1962)
3) IV administration without adverse effects (Benowitz et al. 1983)
4) No effect after 1800 mg/day orally (Bowman & McKennis 1962)
5) No effect with 20 mg IV (Beckett et al. 1972)
6) No cardiovascular effects with 18 mg IV (Benowitz et al. 1983)
7) No effect with 20 mg orally (DeSchepper et al. 1987)
8) No effect with 18 mg IV (Scherer et al. 1988)
9) No effect with 20 mg oral or IV (Curvall et al. 1990)
10) Increased restlessness, tension/anxiety with 30 mg IV (Keenan et al. 1994)

OTHER KNOWN EFFECTS

1) Increased glucose-induced insulin secretion (Tjalve & Popov 1973)
2) Transient increased hepatic drug metabolism (Kyerematen et al. 1983)
3) Decreased endogenous estrogen concentration (Barbieri et al. 1986)
4) Impaired adrenal steroidogenesis (Barbieri et al. 1987; 1989)
5) Decreased testosterone levels (Yeh et al. 1989; Patterson et al. 1990)

BEHAVIORAL EFFECTS: ANIMAL

1) Transiently increased arousal in cats (Yamamoto & Domino 1965)
2) Induces biochemical and behavior changes in rats (Essman 1973)
3) Increased FI response rates in monkeys (Risner et al. 1985)
4) Nicotine-like discrimination in monkeys (Takada et al. 1989)
5) Increased FI response rates in rats (Goldberg et al. 1989)
6) Nicotine-like discrimination in rats (Goldberg et al. 1989)
7) Mecamylamine without effect on behavior (Goldberg et al. 1989)

BEHAVIORAL EFFECTS: HUMAN

1) No effect after 1800 mg/day orally (Bowman & McKennis 1962)
2) No effect with 20 mg IV (Beckett et al. 1972)
3) Decreased withdrawal symptoms with 18 mg IV (Benowitz et al. 1983)
4) No effect with 20 mg orally (DeSchepper et al. 1987)
5) No effect with 18 mg IV (Scherer et al. 1988)
6) No effect with 20 mg oral or IV (Curvall et al. 1990)

Relevant studies are attached .

References

Abood, L.G., Reynolds, D.T., Booth, H., & Bidlack, J.M. (1981). Sites and mechanisms for nicotine's action in the brain. *Neuroscience and Biobehavioral Reviews, 5,* 479-486.

Barbieri, R.L., York, C.M., Cherry, M.L., & Ryan, K.T. (1987). The effects of nicotine, cotinine, and anabasine on rat adrenal 11B-hydroxylase and 21-hydroxylase. *Journal of Steroid Biochemistry, 28,* 25-28.

Beckett, A.H., Gorrod, J.W., & Jenner, P. (1972). A possible relationship between pKa1 and lipid solubility and the amounts excreted in urine of some tobacco alkaloids given to man. *Journal of Pharmacy and Pharmacology, 24,* 115-120.

Benowitz, N.L., Kuyt, F., Jacob, P., Jones, R.T., & Osman, A.L. (1983). Cotinine disposition and effects. *Clinical Pharmacology and Therapeutics, 34,* 604-611.

Benowitz, N.L., & Sharp, D.S. (1989). Inverse relation between serum cotinine concentration and blood pressure in cigarette smokers. *Circulation, 80,* 1309-1312.

Borzelleca, J.F., Bowman, E.R., & McKennis, H. (1962). Studies on the respiratory and cardiovascular effects of (-)-cotinine. *Journal of Pharmacology and Experimental Therapeutics, 137,* 313-318.

Bowman, E.R., Turnbull, L.B., & McKennis, H. (1959). Metabolism of nicotine in the human and excretion of pyridine compounds by smokers. *Journal of Pharmacology and Experimental Therapeutics*, 127, 92-95.

Bowman, E.R., & McKennis, H. (1962). Studies on the metabolism of (-)-cotinine in the human. *Journal of Pharmacology and Experimental Therapeutics*, 135, 306-311.

Bowman, E.R., Hansson, E., Turnbull, L.B., McKennis, H., & Schmiterlow, C.G. (1964). Disposition and fate of (-)-cotinine-[H3] in the mouse. *Journal of Pharmacology and Experimental Therapeutics*, 143, 301-308.

Carroll, M.E., Lac, S.T., Asencio, M., & Keenan, R.M. (1989). Nicotine dependence in rats. *Life Sciences, 45,* 1381-1388.

Curvall, M., Elwin, C.E., Kazemi-Vala, E., Warholm, C., & Enzell, D.R. (1990). The pharmacokinetics of cotinine in plasma and saliva from non-smoking healthy volunteers. *European Journal of Clinical Pharmacology, 38*, 281-287.

Dawson, D.A., Fort, D.J., Smoth, G.J,, Newell, D.L., & Bantle, J.A. (1988). Evaluation of the developmental toxicity of nicotine and cotinine with frog embryo teratogenesis assay: Xenopus. *Teratogenesis, Carcinogenesis and Mutagenesis, 8,* 329-338.

DeSchepper, P.J., VanHecken, A., Daenens, P., & VanRossum, J.M. (1987). Kinetics of cotinine after oral and intravenous administration to man. *European Journal of Clinical Pharmacology, 31,* 583-588.

Dominiak, P., Fuch, G., VonToth, S., & Grobecker, H. (1985). Effects of nicotine and its major metabolites on blood pressure in anesthetized rats. *Klinische Wochenschrift, 63,* 90-92.

Essman, W.B. (1973). Nicotine-related neurochemical changes: Some implications for motivational mechanisms and differences. In: Dunn, Jr., W.L. (ed.), *Smoking Behavior: Motives and Incentives.* V.H. Winston and sons; Washington, D.C., pps. 51-65.

Fuxe, K., Everett, B.J., & Hokfelt, T. (1979). On the action of nicotine and cotinine on central 5-hydroxytryptamine neurons. *Pharmacology, Biochemistry and Behavior, 10,* 671-677.

Goldberg, S.R., Risner, M.E., Stolerman, I.P., Reavill, C., & Garcha, H.S. (1989). Nicotine and some related compounds: Effects on schedule-controlled behavior and discriminative properties in rats. *Psychopharmacology, 97,* 295-302.

Hatsukami, DK, Grillo, M, Pentel, PR, Oncken, C, Bliss, R. (1997) Safety of cotinine in humans: physiologic, subjective and cognitive effects. *Pharmacology Biochemistry and Behavior*, 57, 643-650.

Keenan, R.M., Hatsukami, D.K., Pentel, P., Thompson, T., & Grillo, M.A. (1994). Pharmacodynamic effects of cotinine in abstinent cigarette smokers. *Clinical Pharmacology and Therapeutics, 55*, 581-590.

Keenan, R.M., MacMillan, MD, Hatsukami, D.K. (1995a). The pharmacology of cotinine. *Unpublished book chapter.* Confidential

Keenan, R.M., Hatsukami, D.K., Pentel, P., Thompson, T., & Grillo, M.A. (1995b). Reply: To the Mr. Foulds. *Clinical Pharmacology and Therapeutics, 56,* 95-97.

Kim, K.S., Borzelleca, J.F., Bowman, E.R., & McKennis, H. (1968). Effects of some nicotine metabolites and related compounds on isolated smooth muscle. *Journal of Pharmacology and Experimental Therapeutics, 161*, 59-69.

Kyerematen, G.A., Passananti, G.T., Greene, F.E., & Vesell, E.S. (1983). Time-dependent induction of hepatic drug metabolism in rats by cotinine. *Life Sciences, 32*, 551-556.

LaVoie, E.J., Shigematsu, A., Riernson, A., Mu, B., & Hoffmann, D. (1985). Evaluation of the effects of cotinine and nicotine-N'-oxides on the development of tumors in rats initiated with N-[4-(5-Nitro-2-furyl)-2-thiaozolyl] formamide. *Journal of the National Cancer Institute, 75*, 1075-1081.

McKennis, H., Turnbull, L.B., & Bowman, E.R. (1963). N-methylation of nicotine and cotinine in vivo. *Journal of Biological Chemistry, 238*, 719-723.

Patterson, T.R., Stringham, J.D., & Meikle, A.W. (1990). Nicotine and cotinine inhibit steroidogenesis in mouse Leydig cells. *Life Sciences, 46*, 265-272.

Rahn, C.A., Howard, G., Riccio, E., & Doolittle, D. (1991). Correlations between urinary nicotine and cotinine and urinary mutagenicity in smokers on controlled diets. *Environmental and Molecular Mutagenesis, 17*, 244-252.

Riebe, M., Westphal, K., & Fortnagel, P. (1981). Mutagenicity testing in bacterial tests systems of some constituents of tobacco. *Mutation Research, 101*, 39-43.

Risner, M.E., Goldberg, S.R., Prada, J.A., & Cone, E.J. (1985). Effects of nicotine, cocaine, and some of their metabolites on schedule-controlled responding by beagle dogs and squirrel monkeys. *Journal of Pharmacology and Experimental Therapeutics, 234*, 113-119.

Scherer, G., Jarczyk, L., Heller, W.D., Biber, A., Neurath, G,B., & Adlkofer, F. (1988). Pharmacokinetics of nicotine, cotinine and 3'-hydroxycotinine in cigarette smokers. *Klinische Wochenschrift, 66(Supplement XI)*, 5-11.

Schmähl, D., & Osswald, H, (1968). The absence of of a carcinogenic action of cotinine on rats. *Z Krebsforsch, 71*, 198.

Takada, K., Swedberg, M.D., Goldberg, S.R., & Katz, J.L. (1989). Discriminative stimulus effects of intravenous l-nicotine and nicotine analogs or metabolites in squirrel monkeys. *Psychopharmacology, 99*, 208-212.

Tjälve, H., & Popov, D. (1973). Effect of nicotine and nicotine metabolites on insulin secretion from rabbit pancreas pieces. *Endocrinology, 92*, 1343-1348.

Yamamoto, K.-I., & Domino, E.F. (1965). Nicotine-induced EEG and behavioral arousal. *International Journal of Neuropharmacology, 4*, 359-373.

Yeh, J., Barbieri, R.J., & Friedman, A.J. (1989). Nicotine and cotinine inhibit rat testes androgen biosynthesis in vitro. *Journal of Steroid Biochemistry, 33*, 627-630.

(D) Need for a Warning or Other Limitations on Use. There is no evidence, from any animal study, clinical study or the medical literature, of any need for (i) any warning or caution on the label or other labeling of any dietary supplement due to the inclusion of Cotinine as a dietary ingredient and (ii) any limitation with respect to the use of that ingredient in a dietary supplement (Tab 6).

Please contact the following legal counsel if you have any questions or comments. Thank you for your time and attention to this important matter.

Legal counsel's name and other pertinent information is:

Jim Prochnow, Greenberg Traurig, LLP
The Tabor Center
1200 17th Street, Suite 2400
Denver, CO 80202
T: 303-572-6546
F: 720-904-7646
E: prochnowj@gtlaw.com

Stage II Enterprises, LLC

By: _____
Robert W. Moore
Manager

February 3, 2004